Review



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# The Impact of Genetics on Pediatric Interstitial Lung Diseases: A Narrative Literature Review

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## Abstract

**Background**: Interstitial lung diseases (ILDs) are a heterogeneous group of disorders characterized by variable degrees of inflammation and fibrosis affecting the pulmonary interstitium. Advances in molecular biology and genetics have greatly expanded our understanding of ILD pathogenesis, revealing novel mechanisms and supporting the development of precision medicine approaches. **Genetic insights:** Genetic factors play a pivotal role in ILD heterogeneity, influencing disease onset, severity, and progression. To date, more than 30 genes with different inheritance patterns (autosomal dominant, recessive, or X-linked) have been associated to ILDs. These genes are primarily involved in surfactant metabolism, telomere maintenance, immune regulation, and epithelial repair. Recent evidence also implicates genes encoding aminoacyl-tRNA synthetases. This review summarises the main genetic alterations underlying ILD pathogenesis and discusses their impact on diagnostic and therapeutic approaches, highlighting how identification of disease-causing variants can improve diagnostic accuracy, refine prognostic assessment, and inform recurrence risk. **Methods:** A narrative review was conducted through targeted PubMed and Embase searches using disease- and gene-related keywords. Eligible publications included narrative and systematic reviews, meta-analyses, case series, prospective studies, society guidelines, and peer-reviewed editorials. **Conclusions:** This synthesis brings together the latest genetic insights into pediatric ILDs and their clinical implications. Integrating genomic data into clinical practice may enable earlier diagnosis, tailored follow-up, individualized therapeutic strategies, and more informed genetic counseling. Collectively, these advances hold promise for better outcomes in children affected by ILDs.

**Keywords:** pediatric interstitial lung diseases; next-generation sequencing; genotype–phenotype correlation; pulmonary fibrosis

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## 1. Introduction

### *1.1. Brief Overview of Pediatric Interstitial Lung Diseases (ILDs)*

Interstitial lung diseases (ILDs) represent a broad spectrum of pulmonary disorders characterized by varying degrees of inflammation and fibrosis affecting the lung interstitium, ultimately leading to progressive dyspnea and, in many cases, end-stage respiratory failure [1].

Although ILDs are well studied in adults, pediatric forms represent a distinct subgroup, differing in etiology, pathophysiology, and clinical presentation due to the developmental dynamics of the growing lung. In pediatrics, the acronym chILD (children's ILD) is widely used to distinguish these disorders from ILD in adults.

ILDs can be categorized according to etiology, encompassing connective tissue disease–associated ILD (CTD-ILD), hypersensitivity pneumonitis, drug- or toxin-induced ILD, post-infectious interstitial disease, and idiopathic interstitial pneumonias. Although these disorders often present with similar clinical features, they are defined by distinct histopathologic patterns and prognoses [1].

With more than 200 recognized conditions within the chILD spectrum, substantial discussion continues over how these disorders should be classified—whether they are best grouped into broader categories or separated into more narrowly defined, specific entities. Supporters of a unified approach propose that chILD represent varying manifestations of abnormal airway, mesenchymal, or pulmonary vascular development, with some forms emerging through a "second-hit" mechanism, in which a genetically or structurally susceptible lung develops disease following an additional insult, such as infection. Conversely, proponents of a more granular classification emphasize the importance of defining discrete, genetically determined disease entities [2].

Traditionally, classification frameworks have separated disorders according to age. The framework proposed by Nathan et al. [3] emphasizes patient age as a key determinant, recognizing that ongoing lung growth and maturation substantially influence the molecular and cellular mechanisms of disease. This approach identifies two broad groups: ILDs specific to infancy (<2 years) (such as neuroendocrine cell hyperplasia of infancy (NEHI), pulmonary interstitial glycogenosis, and developmental disorders) and non–age-specific pediatric ILDs that include immune-mediated environmental, and parenchymal dysfunctions.

However, more analyses, such as that by Cunningham et al. [2], suggest that the age-based classification, while conceptually useful, is increasingly limited in practice, particularly in relation to an overlapping age range for genetically diagnosed conditions.

In essence, the Nathan et al. model underscores developmental stage as a clinical guide, whereas the Cunningham et al. framework reflects a paradigm shift toward genotype-driven and pathophysiology-based classification, advocating for integrated models that harmonize genetic, radiological, and clinical dimensions beyond chronological age.

In adults, early classifications were primarily histopathological, identifying four subgroups: usual interstitial pneumonia, desquamative interstitial pneumonia (DIP) and its related form, respiratory bronchiolitis–associated ILD, acute interstitial pneumonia (formerly Hamman–Rich syndrome), and non-specific interstitial pneumonia (NSIP). Over time, a multidisciplinary approach replaced this histopathologic model, and specific entities were validated through the international ATS/ERS consensus statements on idiopathic interstitial pneumonias. In chILD, initial classifications were similarly adapted from adult systems, relying heavily on histopathologic descriptions. Progressively, a more specific, multidisciplinary approach was established that integrates age, clinical context, biological findings, HRCT (High-Resolution Computed Tomography) data, and lung tissue analysis [3].

Current experience underscores the need for continued refinement and harmonization of classification systems to better incorporate the expanding spectrum of genetic mechanisms [2].

Furthermore, recent evidence highlights that ILDs may also develop in immunocompromised children, especially those with primary or secondary immunodeficiencies. In a large multicenter

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cohort, nearly 40% of immunocompromised pediatric patients exhibited interstitial changes associated with underlying genetic or immunologic abnormalities [4].

In summary, pediatric interstitial lung diseases (chILD) represent a complex and evolving spectrum of disorders whose understanding has greatly advanced thanks to multidisciplinary collaboration and genetic research. Despite these advances, diagnostic challenges persist, particularly in distinguishing developmental, immune-mediated, and environmental forms, which often overlap clinically and radiologically. Strengthening genetic and molecular approaches, together with standardized international collaborations, will be crucial to harmonize definitions, improve diagnostic accuracy, and tailor therapeutic strategies. Ultimately, the integration of clinical, radiological, and genomic perspectives offers the most promising pathway toward precision medicine and improved long-term outcomes for children affected by ILDs.

### *1.2. Epidemiology and Clinical Relevance*

The prevalence of chILD remains difficult to quantify and is likely underestimated due to inconsistent definitions, heterogeneous diagnostic criteria, and absence of standardized reporting systems. Estimated prevalence reported in several studies showed large variations that range from 0.1 to 16.2 cases per 100,000. These numbers are difficult to ascertain and are likely underestimated [5].

The highest frequency is observed in younger patients, with a male predominance. Familial clustering occurs in approximately 10% of cases [3].

Regarding etiology, the most frequent causes in children under 2 years of age were surfactant metabolism disorders (16.3%) and NEHI (11.8%), whereas among children aged 2–18 years, the main causes were diffuse alveolar hemorrhage (12.2%), connective tissue disease–associated ILD (11.4%), hypersensitivity pneumonitis (8.8%), and sarcoidosis (8.8%) [6].

These multicenter data underscore that chILD may not be as rare as previously assumed, emphasizing the need for internationally harmonized registries and multicenter collaboration to improve diagnosis, treatment, and prognosis stratification.

### *1.3. Importance of Genetics in Pediatric ILDs*

Genetic mechanisms play a crucial role in determining the onset, severity, and prognosis of pediatric interstitial lung diseases. Over the past two decades, advances in molecular genetics have identified genes involved in ILD pathogenesis, encompassing pathways related to surfactant metabolism, telomere maintenance, immune regulation, and epithelial repair [1].

Growing evidence indicates that both genetic and epigenetic factors contribute to ILD development, with genetic predisposition representing a major risk factor, particularly evident in idiopathic pulmonary fibrosis (IPF). Recent findings also indicate that epigenetic regulatory mechanisms—such as DNA methylation, histone modifications, or non-coding RNAs including microRNAs—play an important role in disease pathogenesis. Environmental and host co-morbidity factors are also important disease contributors. Even in familial ILD with documented genetic causes, environmental risk factors can influence disease expression and progression. Current evidence highlights the role of viruses, tobacco smoke, and exposure to airborne contaminants in modulating disease onset and severity [3].

A recent analysis from the chILD-EU registry demonstrated that early genetic testing aimed at identifying the underlying cause of respiratory dysfunction in newborns can often obviate the need for video-assisted thoracoscopic surgery (VATS) biopsy, particularly in cases involving surfactant gene mutations or alveolocapillary dysplasia with misalignment of the pulmonary veins. Therefore, genetic testing should be considered as early as possible in the diagnostic work-up of chILD in neonates [7].

Moreover, the incorporation of genetic testing within multidisciplinary diagnostic discussions (MDD) has become crucial for accurately interpreting variants and establishing meaningful genotype–phenotype correlations [8].

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Next-generation sequencing approaches, especially trio-based whole-exome or whole-genome sequencing, significantly enhance the ability to identify disease-causing genes and uncover new variants. Although autosomal dominant conditions with incomplete penetrance complicate segregation analysis, as asymptomatic relatives may still carry pathogenic variants, genetically supported diagnoses remain highly valuable. In particular, obtaining a molecular diagnosis can guide timely clinical decisions and may obviate the need for invasive lung biopsy, especially in cases where lung transplantation or palliative strategies must be considered [2].

Ultimately, the growing role of genetics is redefining pediatric ILD as a model for precision medicine in rare respiratory diseases—where early molecular diagnosis can guide individualized care, optimize outcomes, and inform preventive strategies for affected families.

### *1.4. Rationale and Scope of the Narrative Review*

The rationale of this review is to provide a critical overview of the current knowledge regarding genetic determinants of chILD, emphasizing how these insights are reshaping diagnostic algorithms, refining prognostic stratification, and informing individualized management strategies. This narrative review draws upon recent studies, systematic reviews, and expert consensus to outline the evolving role of genetics in defining disease classification, improving early recognition, and supporting family counseling. Ultimately, this synthesis aims to bridge the gap between molecular discovery and clinical application, fostering a precision-medicine approach that can improve outcomes and quality of care for affected children and their families.

## 2. Genetic Basis of Pediatric ILDs

Among chILD disorders, distinguishing between isolated and syndromic forms is essential. This classification provides clinicians with a practical approach to evaluating children with diffuse lung disease and determining whether additional systemic manifestations are present (**Table 1**).

**Table 1.** Genes with established associations to isolated and syndromic forms of chILD.

| | Functional Impact | Gene | Inheritance | Onset | Related Phenotype | ILD Key Features |
|---|---|---|---|---|---|---|
| **Isolated chILDs** | **Surfactant-related disorders** | *SFTPC* | AD | Neonatal/infancy | Surfactant metabolism dysfunction, pulmonary, 2 | Ground-glass pattern, Alveolar proteinosis |
| | | *SFTPB* | AR | Neonatal | Surfactant metabolism dysfunction, pulmonary, 1 | Ground-glass pattern, Alveolar proteinosis, Minimal exogenous surfactant response |
| | | *ABCA3* | AR | Neonatal | Surfactant Metabolism Dysfunction, Pulmonary, 3 | Ground-glass pattern, Progressive fibrosis Lamellar body deposition, |
| | | *SFTPA1*/*SFTPA2* | AD | Adult/rare pediatric | Interstitial Lung Disease 1, 2 | Pulmonary fibrosis, Early lung cancer risk |
| | **GM-CSF receptor-related disorders** | *CSF2RA* | PAR | Childhood | Surfactant metabolism dysfunction, pulmonary, 4 | Ground-glass pattern, Defective alveolar macrophage maturation |

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| | | | | | | |
|---|---|---|---|---|---|---|
| | | *CSF2RB* | AR | Adult/rare pediatric | Surfactant metabolism dysfunction, pulmonary, 5 | Ground-glass pattern, Defective alveolar macrophage maturation |
| | **Lysosomal-related disorders** | *LAMP3* | AR | Childhood | – | Ground-glass pattern, Pulmonary fibrosis, Alveolar cell hyperplasia |
| **Syndromic chILDs** | **Telomere maintenance** | *TERT/TERC* | AD | Childhood-adult | Pulmonary fibrosis and/or bone marrow failure syndrome, telomere-related 1, 2 | Pulmonary fibrosis |
| | | *NOP10/TINF2* | AR/AD | Adolescence/ childhood-adult | Dyskeratosis congenita, autosomal recessive 1, autosomal dominant 3 | Pulmonary fibrosis |
| | | *DKC1* | X-linked | Childhood-adolescence | Dyskeratosis congenita, X-linked | Pulmonary fibrosis |
| | **Immune disordes/ interferonopathies** | *STING1* | AD | Childhood | STING-associated vasculopathy with onset in infancy | Ground-glass pattern, Pulmonary fibrosis, Alveolar macrophage infiltration |
| | | *COPA* | AD | Adolescence-young adult | Autoinflammation and autoimmunity, systemic, with immune dysregulation 1 | Ground-glass pattern, Lung cysts, Alveolar hemorrhage |
| | | *STAT5B* | AR | Childhood | Growth hormone insensitivity with immune dysregulation 1 | Pulmonary fibrosis |
| | | *OAS1* | AD | Neonatal | Immunodeficiency 100 with pulmonary alveolar proteinosis and hypogammaglobulinemia | Lung consolidations, Alveolar proteinosis |
| | | *CCR2* | AR | Childhood | Polycystic lung disease | Lung cysts, Alveolar proteinosis, Mild interstitial fibrosis |
| | **Cytoskeletal / structural disorganization** | *ITGA3* | AR | Neonatal | Epidermolysis Bullosa Junctional 7 with interstitial lung disease and nephrotic syndrome | Interstitiophaty |
| | **Pulmonary development / transcriptional dysregulation** | *NKX2-1* | AD | Neonatal | Choreoathetosis, hypothyroidism, and neonatal respiratory distress | Interstitiophaty, Pulmonary fibrosis |
| | | *TBX4* | AD | Childhood | Ischiocoxopodopatellar syndrome, with or without pulmonary arterial hypertension | Acinar dysplasia |

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| | | | | | |
|---|---|---|---|---|---|
| | *FARSA/FARSB* | AR | Childhood | Rajab interstitial lung disease with brain calcifications 1, 2 | Pulmonary Fibrosis, Alveolar proteinosis |
| **Aminoacyl-tRNA synthetase disorders** | *MARS1* | AR | Childhood | Interstitial lung and liver disease | Pulmonary Fibrosis, Alveolar proteinosis |
| | *YARS1* | AR | Childhood | Infantile-onset multisystem neurologic, endocrine, and pancreatic disease 2 | Lung cysts, Pulmonary Fibrosis |
| | *NHLRC2* | AR | Childhood | FINCA syndrome | Pulmonary Fibrosis |
| **Multisystem fibrosing syndromes** | *FAM111B* | AD | Childhood | Poikiloderma, hereditary fibrosing, with tendon contractures, myopathy, and pulmonary fibrosis | Pulmonary Fibrosis |

*2.1. Isolated chILDs*

These clinical conditions involve the lungs predominantly or exclusively. They typically affect the alveolar epithelium, surfactant metabolism, lipid processing, lysosomal function or alveolar macrophage biology [9].

2.1.1. Genes Associated with Surfactant-Related Disorders

- *SFTPC* (OMIM *178620) encodes pulmonary-associated surfactant protein C (SPC) and, thus, it is associated with *Surfactant metabolism dysfunction, pulmonary, 2* (OMIM #610913), an autosomal dominant disorder with an early onset in full-term newborns. SFTPC-related disorder shows reduced penetrance and variable expressivity, and clinical severity may be exacerbated by recurrent respiratory infections. Affected individuals commonly exhibit failure to thrive, digital clubbing, hypoxemia, and respiratory distress with tachypnea or dyspnea. Thoracic CT scan typically reveals ground-glass opacities with interlobular septal thickening [10]. Histopathologic findings often include intra-alveolar accumulation of abnormal pro-SPC protein, type II pneumocyte hyperplasia and features of alveolar proteinosis [11–13].

- *SFTPB* (OMIM *178640) encodes surfactant protein B (SPB) and is associated with *Surfactant metabolism dysfunction, pulmonary, 1* (OMIM #265120), an autosomal recessive disorder characterized by neonatal onset and rapid clinical deterioration. Affected infants typically present with severe respiratory distress, tachypnea/dyspnea, pulmonary hypertension, and failure to thrive. Chest imaging commonly shows interstitial thickening and diffuse ground-glass opacities, while histopathology reveals alveolar proteinosis. A minimal or transient response to exogenous surfactant therapy may be observed, and death often occurs within the first months of life. Survival beyond infancy is exceedingly rare [14,15].

- *SFTPA1* (OMIM 178630) and *SFTPA2* (OMIM 178642) encode components of the lung surfactant protein A complex. Their associated phenotypes, *Interstitial Lung Disease 1* (OMIM #619611) and *Interstitial Lung Disease 2* (OMIM #178500), show incomplete penetrance, variable expressivity, and typically present in adulthood. Therefore, they are not primarily involved in chILD, although pediatric cases have been reported on rare occasions. These two conditions usually present with indolent interstitial pneumonia and recurrent respiratory infections; in some families, pulmonary fibrosis co-segregates with early-onset lung cancer [16].

- *ABCA3* (OMIM *601615) is expressed in alveolar type II pneumocytes and plays an essential role in surfactant synthesis. Pathogenic variants in this gene cause *Surfactant Metabolism Dysfunction, Pulmonary, 3* (OMIM #610921), an autosomal recessive disorder typically presenting with severe respiratory distress in newborns, often accompanied by tachypnea/dyspnea and failure to thrive.

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Biallelic loss-of-function variants usually result in lethal neonatal respiratory failure, whereas hypomorphic or missense variants may allow survival beyond infancy and lead to a chronic form of ILD. Chest CT generally shows ground-glass opacities and evolving fibrosis, and lung biopsy frequently reveals abnormal lamellar bodies within type II pneumocytes [17–20].

#### 2.1.2. Genes Associated with GM-CSF Receptor–Related Disorders

- *CSF2RA* (OMIM *306250) and *CSF2RB* (OMIM *138981) encode, respectively, the ligand-binding alpha subunit and the non–ligand-binding, affinity-enhancing beta subunit of the granulocyte–macrophage colony-stimulating factor (GM-CSF) receptor. *CSF2RA*, located in the pseudoautosomal region (PAR) of the X chromosome [21] is associated with *Surfactant metabolism dysfunction, pulmonary, 4* (OMIM #300770), a congenital pseudoautosomal recessive disorder characterized by failure to thrive, tachypnea, restrictive ventilatory impairment, and patchy ground-glass opacities on chest CT [22]. *CSF2RB* is associated with *Surfactant metabolism dysfunction, pulmonary, 5* (OMIM #614370), a recessive condition more commonly presenting in adulthood, although pediatric cases have been reported. Affected individuals develop respiratory insufficiency with dyspnea, alveolar proteinosis, and ground-glass abnormalities on imaging [23]. In both disorders, defective maturation of alveolar macrophages leads to impaired surfactant clearance and accumulation of surfactant-rich material within the alveoli.

#### 2.1.3. Genes Linked to Disorders of Epithelial Stress Response, Autophagy, and Lysosomal Function

- *MUC5B* (OMIM *600770) encodes a secreted gel-forming mucin expressed in the airways. The common promoter variant is strongly associated with *Susceptibility to idiopathic pulmonary fibrosis* (OMIM #178500). Although its contribution to childhood-onset interstitial lung disease is considered rare, it is well recognized as a genetic modifier in adult fibrotic lung disease [24].

- *GRN* (OMIM *138945) encodes progranulin, an autocrine growth factor involved in lysosomal function, cellular stress responses, and immune regulation. A recent pediatric case report described a homozygous loss-of-function variant associated with early-onset parenchymal lung disease, presenting with an alveolar proteinosis–like pattern and features of chronic pulmonary inflammation [25].

- *HMOX1* (OMIM *141250) encodes heme oxygenase and has been implicated in susceptibility to *chronic obstructive pulmonary disease* (OMIM %606963). A recent case report described a young boy presenting with chronic respiratory failure due to nonspecific interstitial pneumonia, following recurrent infection-triggered hyperinflammatory episodes, including hemolysis [26].

- *LAMP3* (OMIM *605883) encodes lysosome-associated membrane glycoprotein 3, a protein involved in the regulation of lysosomal function and surfactant processing. Very recently, biallelic variants in LAMP3 have been associated with heterogeneous pediatric interstitial lung disease, including surfactant-related pulmonary manifestations [27].

### *2.2. Syndromic chILDs*

The following syndromic forms are multisystem disorders in which ILD constitutes one component of a broader phenotype. The underlying defects frequently involve fundamental cellular pathways, including telomere maintenance, immune regulation, cytoskeletal organization, protein translation and embryonic lung development.

#### 2.2.1. Genes Associated with Telomere Maintenance Disorders

- *PARN* (OMIM *604212) encodes an exoribonuclease that shortens mRNA poly(A) tails through deadenylation, thereby regulating gene expression [28]. Pathogenic variants in *PARN* are associated with *Pulmonary fibrosis and/or bone marrow failure syndrome, telomere-related, 4* (OMIM #616371), an autosomal dominant condition characterized by variable expressivity, incomplete penetrance and typically adult onset. Affected individuals most commonly present with pulmonary fibrosis and may

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also exhibit premature graying of the hair. PARN-related features have also been reported in a child with ILD, who showed marked shortening of lymphocyte telomere length [29].

- *TERC* (OMIM *602322) and *TERT* (OMIM *187270) encode essential components of the telomerase complex, the reverse transcriptase machinery responsible for adding repetitive DNA sequences to telomeres [30,31]. Pathogenic variants in *TERC* and *TERT* are associated, respectively, with *Pulmonary fibrosis and/or bone marrow failure syndrome, telomere-related, 2* (OMIM #614743) and *Pulmonary fibrosis and/or bone marrow failure syndrome, telomere-related, 1* (OMIM #614742). Both conditions follow an autosomal dominant inheritance pattern and demonstrate variable expressivity, incomplete penetrance, and typically adult onset. Clinical manifestations include pulmonary fibrosis, premature graying of the hair, bone marrow failure or pancytopenia, and an increased risk of cancer, leukemia, or myelodysplastic syndrome. Although predominately described in adults, pediatric cases have also been reported [32].

- *TINF2* (OMIM *604319) encodes a core component of the 6-protein shelterin/telosome complex. Heterozygous pathogenic variants in *TINF2* are associated with *Dyskeratosis congenita, autosomal dominant 3* (OMIM #613990), a multisystemic condition characterized by variable onset from childhood to adulthood. Reported clinical manifestations include short stature, intrauterine growth restriction (IUGR), microcephaly, deafness, ocular abnormalities (such as nasolacrimal duct obstruction, epiphora, and retinopathy), oral leukoplakia, premature tooth loss, pulmonary fibrosis, respiratory insufficiency, enteropathy, portal hypertension, cryptorchidism, osteoporosis, reticular skin pigmentation, nail dystrophy, premature graying of the hair, alopecia, speech and learning difficulties, cerebellar hypoplasia or ataxia, bone marrow failure or pancytopenia, and an increased risk of cancer, leukemia, or myelodysplastic syndrome. Patients typically demonstrate marked telomere shortening and reduced telomerase activity, consistent with the broader spectrum of telomere-biology disorders [33,34].

- *NOP10* (OMIM *606471) encodes a protein component of the H/ACA small nucleolar ribonucleoprotein (snoRNP) complex. Pathogenic variants in NOP10 are associated with two distinct clinical entities: *Dyskeratosis congenita, autosomal recessive 1* (OMIM #224230), and an autosomal dominant form of *Pulmonary fibrosis and/or bone marrow failure syndrome, telomere-related, 9* (OMIM #620400). The autosomal recessive subtype (DKCB1) typically presents during adolescence and is characterized by nail dystrophy, skin hyperpigmentation, mucosal leukoplakia, neurodevelopmental disorders, bone marrow failure, lung and liver fibrosis, microcephaly and an increased risk of malignancies [35]. In contrast, the autosomal dominant form (PFBMFT9) generally exhibits adult onset, variable expressivity, incomplete penetrance and notably shows genetic anticipation [36].

- *POT1* (OMIM *606478) encodes a protein that binds the G-rich strand of telomeric DNA and plays a critical protective role in telomere maintenance. Pathogenic variants in POT1 are associated with an autosomal dominant form of *Pulmonary fibrosis and/or bone marrow failure syndrome, telomere-related, 8* (OMIM #620367). This condition typically shows adult onset, variable extrapulmonary manifestations, incomplete penetrance, and may exhibit genetic anticipation [37].

- *DKC1* (OMIM *300126) encodes dyskerin, a core component of the telomerase complex and H/ACA ribonucleoprotein particles. Pathogenic variants in DKC1 cause *Dyskeratosis congenita, X-linked* (OMIM #305000), a multisystem disorder usually presenting from childhood to adolescence. Reported clinical features include short stature, intrauterine growth restriction (IUGR), microcephaly, ocular abnormalities, oral leukoplakia, premature tooth loss, pulmonary fibrosis, restrictive lung disease, gastrointestinal disturbances, hypospadias, phimosis or cryptorchidism, osteoporosis, reticular skin pigmentation, nail dystrophy, premature graying of the hair, neurodevelopmental abnormalities, cerebellar hypoplasia or ataxia, bone marrow failure, increased susceptibility to malignancies (including pancreatic and squamous cell carcinoma), and a heightened predisposition to chromosomal instability [38,39].

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2.2.2. Genes Implicated in Immune Function and Interferon Signaling

- *STING1/TMEM173* (OMIM *612374) encodes a transmembrane protein localized to the endoplasmic reticulum that plays a key role in type I interferon production, particularly interferon-β. Pathogenic variants in this gene are associated with *STING-associated vasculopathy with onset in infancy* (SAVI, OMIM #615934), an autosomal dominant condition. Affected individuals typically present with growth retardation, systemic vasculitis accompanied by cutaneous manifestations such as telangiectasia, erythema, or livedo reticularis, recurrent respiratory infections, pulmonary fibrosis with alveolar macrophage infiltration, nail dystrophy, anemia, and various immunological abnormalities [40,41].

- *ISG15* (OMIM *147571) encodes a ubiquitin-like protein induced by type I interferons, especially interferon-alpha. Biallelic pathogenic variants are associated with a pediatric interferonopathy characterized by lung fibrosis, inflammatory cutaneous lesions, interstitial pneumonia, and basal ganglia calcifications [42].

- *COPA* (OMIM *601924) encodes a subunit of the coatomer protein complex I (COPI). Pathogenic variants are associated with *Autoinflammation and autoimmunity, systemic, with immune dysregulation 1* (OMIM #616414), an autosomal dominant condition. Clinical features typically include respiratory symptoms (i.e., dyspnea, cough and hemoptysis), interstitial lung disease, alveolar hemorrhage, lung cysts and ground-glass opacities on CT scans. Hepatic and renal involvement, such as cirrhosis or glomerulonephritis, can also occur. Additional features may include inflammatory arthritis, cutaneous vasculitis, gait disturbances, and various immunological abnormalities. Disease onset usually occurs in the first or second decade of life, with highly variable expressivity and incomplete penetrance [43].

- *STAT5B* (OMIM *604260) encodes a transcription factor involved in cytokine signaling. Biallelic pathogenic variants are associated with *Growth hormone insensitivity with immune dysregulation 1* (OMIM #245590), an autosomal recessive early-onset disorder. Affected individuals present with short stature, due to growth hormone (GH) deficiency, failure to thrive, respiratory insufficiency, lung fibrosis, and a spectrum of endocrine and immune abnormalities [44].

- *STAT3* (OMIM *102582) encodes a member of the STAT protein family involved in transcriptional regulation downstream of multiple cytokines. Monoallelic gain-of-function variants in *STAT3* are typically associated with *Autoimmune disease, multisystem, infantile-onset 1* (OMIM #615952). This early-onset condition is characterized by short stature, autoimmune enteropathy, dermatitis, endocrine abnormalities such as type 1 diabetes mellitus, and diverse immunologic and hematologic disorders [45].

- *GATA2* (OMIM *137295) encodes a hematopoietic transcription factor. Monoallelic pathogenic variants cause *Immunodeficiency 21* (OMIM #614172), an autosomal dominant disorder with broad phenotypic variability and variable age at onset. Pulmonary involvement is frequently observed and may include alveolar proteinosis and progressive interstitial lung disease [7,46].

- *OAS1* (OMIM *164350) encodes an interferon-induced protein belonging to the oligoadenylate synthetase (OAS) family. Heterozygous de novo mutations in OAS1 are associated with *Immunodeficiency 100 with pulmonary alveolar proteinosis and hypogammaglobulinemia* (OMIM #618042), a neonatal-onset condition with poor life expectancy. Clinical features include failure to thrive, respiratory insufficiency, lung consolidations and alveolar proteinosis, gastrointestinal inflammation, cutaneous rashes, and immunologic abnormalities [7].

- *CCR2/MCP-1* (OMIM *601267) encodes a seven-transmembrane G-protein–coupled receptor for the chemokine CCL2. Biallelic pathogenic variants have been associated with a recessive form of *Polycystic lung disease* (OMIM #219600), typically presenting in childhood with progressive clinical evolution. Reported pulmonary manifestations include alveolar proteinosis and mild interstitial fibrosis [47,48].

- *AP3B1* (OMIM *603401) encodes the large B1 subunit of the adaptor-related protein complex-3. Biallelic pathogenic variants in this gene cause *Hermansky–Pudlak syndrome type 2* (OMIM #608233),

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characterized by neutropenia, recurrent infections and, in some reported cases, fibrosing interstitial lung disease [49,50].

2.2.3. Genes Involved in Cytoskeletal Organization and Structural Integrity

- *FLNA* (OMIM *300017) encodes filamin A. Heterozygous variants in this gene have recently been associated with a X-linked form of interstitial lung disease, often accompanied by cardiac involvement and showing a wide range of ages at onset, from the fetal period to young adulthood [51,52].

- *ACTA2* (OMIM * 102620) encodes the smooth muscle $\alpha$-actin isoform. Monoallelic de novo mutations in this gene are associated with *Smooth Muscle Dysfunction Syndrome* (OMIM #613834), which is characterized by congenital mydriasis, retinal small-vessel aneurysms,, cardiovascular defects, gastrointestinal disorders, and interstitial lung disease, often manifesting with tachypnea [53].

- *ITGA3* (OMIM *605025) encodes an integrin belonging to the family of cell surface adhesion molecules. Biallelic mutations in this gene are associated with *Epidermolysis Bullosa Junctional 7 with interstitial lung disease and nephrotic syndrome* (ILNEB, OMIM #614748), a severe condition characterized by poor life expectancy, growth retardation, microcephaly, mild dysmorphisms, respiratory distress due to interstitial lung disease, renal fibrosis, skin involvement, hypotonia and developmental delay [54].

- *ARHGAP42* (OMIM *615936) encodes a protein of the GTPase Regulator Associated with Focal Adhesion Kinase (GRAF) family. Although this gene is not yet linked to a well-defined clinical phenotype, preliminary evidence suggests a possible involvement in lung disease. A recent case report described a young girl presenting with interstitial lung disease, systemic hypertension, immunologic abnormalities carrying a homozygous stop-gain variant in this gene [55]. This observation suggests that loss-of-function variants in *ARHGAP42* may contribute to a broader syndromic spectrum, although additional cases will be necessary to clarify its pathogenic relevance.

2.2.4. Genes Implicated in Embryonic Lung Development and Transcriptional Regulation

- *NKX2-1* (OMIM *600635) encodes a transcription factor essential for the early development of the thyroid, lung, and forebrain. Mutations in this gene are associated with *Choreoathetosis, hypothyroidism, and neonatal respiratory distress* (OMIM #610978), an autosomal dominant condition primarily characterized by congenital hypothyroidism, neurological manifestations (neonatal or early childhood hypotonia evolving into benign hereditary chorea) and pulmonary involvement, which may present as ILD, pulmonary fibrosis, or respiratory distress accompanied by tachypnea, cough, and recurrent airway infections [56,57].

- *TBX4* (OMIM *601719) encodes transcription factor of the T-box family involved in embryonic development of the lungs and skeleton. Mutations in this gene are associated with *Ischiocoxopodopatellar syndrome, with or without pulmonary arterial hypertension* (OMIM #147891), which is characterized by short stature, cleft palate, osteoarticular abnormalities and idiopathic pulmonary hypertension. Over the past few years, the spectrum of TBX4-related pulmonary phenotypes has been expanded, including severe parenchymal lung disorders such as acinar dysplasia [58,59].

- *FOXF1* (OMIM *601089) encodes Forkhead box protein F1, a member of the Forkhead transcription factor family. Monoallelic pathogenic variants are associated with *Alveolar capillary dysplasia with misalignment of pulmonary veins* (OMIM #265380), a neonatal-lethal disorder characterized by cardiovascular defects, gallbladder agenesis, annular pancreas, intestinal malrotation or atresia, urinary tract abnormalities, and severe early-onset pulmonary involvement with poor prognosis [60–62].

- *FGF10* (OMIM *602115) encodes fibroblast growth factor 10, an essential regulator of pulmonary branching morphogenesis. Heterozygous pathogenic variants have been reported in children with early-onset ILD, presenting as postnatal respiratory failure. Histopathology from fatal

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cases reveal diffuse developmental lung alterations, (i.e., acinar dysplasia and interstitial fibrosis), with some patients also developing pulmonary hypertension [63].

2.2.5. Genes Involved in Protein Synthesis and tRNA-Charging Processes

- FARSA (OMIM *602918) and FARSB (OMIM *609690) encode the catalytic α- and β-subunits of cytoplasmic phenylalanine-tRNA synthetase (FARS1). Biallelic pathogenic cause Rajab interstitial lung disease with brain calcifications 1 and 2 (OMIM #613658 and #619013), two multisystemic conditions with high variable expressivity. Clinical manifestations typically include interstitial lung disease, leukoencephalopathy with brain cysts and intracranial calcifications, microcephaly, hepatic dysfunction, hypoalbuminemia, skin and joint hyperlaxity, growth retardation, and distinctive dysmorphic features (elfin-like facies) [64].

- *MARS1* (OMIM *156560) encodes a cytoplasmic methionyl-tRNA synthetase, a member of a class I aminoacyl-tRNA synthetase. Biallelic pathogenic variants are associated with *Interstitial lung and liver disease* (OMIM #615486), typically presenting in infancy with interstitial lung disease characterized by pulmonary fibrosis and, in some cases, alveolar proteinosis, along with progressive liver dysfunction and failure to thrive. The clinical course is heterogeneous and may progress to respiratory failure [65,66].

- *YARS1* (OMIM *603623) encodes the cytoplasmic tyrosyl-tRNA synthetase. Biallelic pathogenic variants cause *Infantile-onset multisystem neurologic, endocrine, and pancreatic disease 2* (OMIM #619418), a condition with variable age at onset and clinical severity. Reported clinical features include neurodevelopmental impairment, poor prenatal and postnatal growth, gastrointestinal and hepatic involvement, chronic anemia, endocrine abnormalities, cystic or fibrotic interstitial lung disease, retinitis pigmentosa, and sensorineural hearing loss [67].

- Biallelic variants in *AARS1* (OMIM *601065), *LARS1* (OMIM *151350), and *IARS1* (OMIM *600709) have been associated with variable interstitial lung involvement, including pulmonary alveolar proteinosis in some cases. These disorders fall within the broader spectrum of recessive cytoplasmic aminoacyl-tRNA synthetase (aARS) deficiencies, which marked phenotypic heterogeneity. Among all cytoplasmic aARS genes, *MARS1* and *FARSA*/*FARSB* deficiencies appear to be more consistently linked to prominent, and in some cases severe, respiratory manifestations [68].

2.2.6. Additional Genes Associated with Fibrosing Multisystem Syndromes Include

- *NHLRC2* (OMIM *618277) encodes a protein implicated in reactive oxygen species (ROS)-induced apoptosis. Biallelic pathogenic variants in this gene cause *FINCA syndrome* (FIbrosis, Neurodegeneration and Cerebral Angiomatosis—OMIM #618278), an infantile multisystem fibrosing disorder in which severe pulmonary fibrosis is a prominent manifestation [69].

- *FAM111B* (OMIM *615584) encodes a trypsin--like cysteine/serine protease. Heterozygous mutations in this gene underlie hereditary fibrosing poikiloderma (OMIM #615704) with tendon contractures and may be associated with progressive pulmonary fibrosis in a subset of patients [70].

2.2.7. Copy Number Variations (CNVs)

Two additional deletion syndromes are relevant in the context of childhood-onset interstitial lung disease:

-Deletion 5q (OMIM #153550) is commonly observed in myelodysplastic syndromes and has been reported in cohorts with associated interstitial lung disease; such chromosomal abnormalities should be considered hematologic predisposition factors for pulmonary complications [71].

- Contiguous *ABCD1*/*DXS1357E* deletion syndrome (CADDS, OMIM #300475) is a neonatal contiguous-gene deletion condition with multisystem involvement (including liver and nervous system) and occasional respiratory manifestations; it is best categorized among contiguous-gene syndromes with secondary pulmonary disease [72].

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In summary, although distinguishing between isolated and syndromic forms of chILD is clinically useful, the boundaries between these categories are often fluid. Some syndromic conditions may initially present with apparently isolated lung disease, whereas primary surfactant disorders may evolve into broader multisystem phenotypes due to chronic inflammation or secondary pulmonary hypertension. Consequently, multidisciplinary evaluation and molecular confirmation are essential to achieve accurate phenotyping, risk stratification, and long-term management.

## 3. Diagnostic Clues

### *3.1. Typical Clinical Presentation in Infancy and Childhood*

The clinical manifestations of pediatric ILD are often subtle and non-specific. Symptom onset is typically insidious, and many children may experience respiratory complaints for years before a definitive diagnosis is established. Presentations range from asymptomatic cases with incidental radiologic abnormalities suggestive of ILD to more characteristic findings marked by respiratory symptoms and recurrent exacerbations [5].

Children with ILD typically present with cough and tachypnoea, or may exhibit unexpectedly severe or persistent respiratory morbidity in the neonatal period or later after an acute viral respiratory infection. Failure to thrive is particularly common in infants and young children. Older children may present with chronic, slowly progressive dyspnea either at rest or with exercise intolerance [2,5].

According to Nayir Buyuksahin H. and Kiper N. [73], children who have at least 3 of the following 4 criteria for a minimum of 4 weeks should undergo evaluation for ILD: (1) respiratory symptoms/signs (such as cough, tachypnea, or dyspnea at rest or with exercise, crackles, retractions, digital clubbing, failure to thrive, or respiratory failure); (2) systemic arterial hypoxemia; (3) diffuse radiological abnormalities; (4) abnormal pulmonary function tests [73].

It is important to emphasize that the timing and severity of clinical presentation can provide valuable diagnostic clues. Severe respiratory distress in the immediate postnatal period often suggests developmental lung disorders or surfactant dysfunction diseases, which are typically associated with high morbidity and limited therapeutic options. In contrast, entities such as pulmonary interstitial glycogenosis (PIG) and NEHI usually present with milder symptoms, although they may still require prolonged supportive care and close follow-up [74].

Cough is a frequent symptom in interstitial lung diseases, with approximately 30%–50% of patients with IPF reporting a cough severe enough to impair quality of life. Dyspnea is the is the hallmark of ILD, and typically first manifests during strenuous exertion. As disease progresses, patients experience gradual limitations in exercise capacity, and resting hypoxemia becomes common in advanced stages. The onset and rate of progression of dyspnea vary across different ILDs. Fatigue and unintentional weight loss may also develop over time, with weight loss serving as an adverse prognostic indicator. Digital clubbing is present in approximately 7%–42% of individuals with pulmonary fibrosis. On lung auscultation, fine Velcro-like crackles at the bases are identified in 93% of patients with IPF and in 73% of those with non-IPF ILDs, while individuals with hypersensitivity pneumonitis may exhibit high-pitched end-inspiratory squawks [1]. Wheezing may also be observed, although up to one-third of children may have normal auscultation. Additional clinical signs include subcostal retractions, chest wall deformity such as pectus excavatum, seen in some surfactant disorders, or pectus carinatum (more typical in airway disorders associated with air-trapping such as NEHI and bronchiolitis obliterans), and hypoxemia. Children may also show clinical features of pulmonary hypertension [2].

### *3.2. Challenges in Diagnosis and Differential Diagnosis*

Given that the prevalence of individual entities is likely <1 per 100,000 individuals, most hospitals encounter so few cases that experience with diagnosis and management is often limited.

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Helpful guidelines outlining clinical assessment, imaging and/or genetic as well as histopathological analyses have been established to support accurate diagnosis [7].

In this section, we summarize the most recent evidence regarding the diagnostic process, focusing on the integration of clinical, radiological, genetic, and histopathological findings that have refined the current approach to the evaluation and classification of chILD.

Before pursuing a specific chILD diagnosis, more common causes of interstitial lung disease in children must be excluded. These include cystic fibrosis, acquired or congenital immunodeficiency, congenital heart disease, bronchopulmonary dysplasia, pulmonary infection, primary ciliary dyskinesia and recurrent aspiration. Clinical evaluation should include a thorough medical history—covering family history, prenatal course, and neonatal events—assessment of oxygen saturation at rest and during exercise, chest imaging (radiography and HRCT), laboratory testing with emphasis on immune function and autoantibodies, echocardiography, and pulmonary function testing (in children aged >4–5 years). Useful clinical protocols are available on the chILD-EU website (www.childeu.net). Echocardiography is essential for excluding structural cardiovascular disease and pulmonary hypertension (PH), which may occur in up to 7% of children evaluated for suspected ILD [7].

Routine laboratory investigations (including complete blood count, serum markers for renal, hepatic, and thyroid function, and quantitative immunoglobulins) are used to screen for systemic or multiorgan disease. Serological testing for autoantibodies is particularly important in older children and in those with evidence of pulmonary hemorrhage, renal involvement, and articular, cutaneous, or systemic manifestations. IgG precipitin testing to allergens may assist in the evaluation of suspected hypersensitivity pneumonitis, though sensitivity and specificity are incomplete (ie, a hypersensitivity to birds).

A stepwise approach involves non-invasive tests and invasive studies if needed, including assessment of oxygenation and ventilation, pulmonary function testing, chest imaging, and consideration of bronchoscopy and, in selected cases, lung biopsy.

Oxygen saturation measured via pulse oximetry (SpO2) reflects pulmonary impairment and is a key component of chILD severity scoring systems. Standardization is essential to ensure clinical reliability.

The chILD-EU group recommends measuring $SpO_2$ in room air, with the child resting for 5 minutes and accepting a desaturation nadir of 80% before resuming supplemental oxygen if required. $SpO_2$ measurement during sleep or dynamic testing (e.g., exercise in older children) may reveal abnormalities not evident at rest. However, inter-center variability in measurement protocols limits comparability across institutions [2].

When ILD is suspected or confirmed, pulmonary function testing (including forced vital capacity [FVC] and diffusing capacity for carbon monoxide [DLCO]) should be obtained to assess disease severity. LD typically results in a restrictive ventilatory pattern on spirometry [1].

The 2013 ATS guidelines propose three diagnostic algorithms, two for infants presenting with severe respiratory disease in the neonatal period (differentiated by the presence or absence of a family history of ILD) and one for children presenting after 1 month of age. In all three algorithms, imaging appears early in the diagnostic pathway, once common non-chILD conditions have been excluded [75].

Even when imaging does not yield a specific chILD diagnosis, it remains a key component of multidisciplinary evaluation. Many diffuse lung diseases demonstrate non-homogeneous lung involvement with geographic areas of sparing or variable disease patterns. Consequently, pre-biopsy imaging is essential to guide the surgeon toward an appropriately affected region of lung tissue and to support the subsequent interpretation of biopsy findings.

Genetic testing has become an increasingly important element of the chILD diagnostic work-up, especially in infants and even more so when there is a strong family history [76].

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Genetic alterations have been reported in association with specific chILD entities in up to 20% of cases. In a recent analysis of the chILD-EU register, 46% of enrolled patients underwent genetic testing, and 13% of these achieved a final molecular diagnosis [7].

The overarching aim of both imaging and genetic studies is to establish the most specific diagnosis possible without resorting to invasive procedures, with diagnostic algorithms converging on surgical lung biopsy only when no alternative etiology can be identified [76].

Thus, the indication for genetic testing should be considered as early as possible in the diagnostic evaluation of neonates with suspected child [7].

### *3.3. Radiography*

Chest radiography remains the most widely used respiratory imaging tool globally, offering a rapid overview of the entire thorax in a single acquisition. However, in both children and adults, its sensitivity for detecting ILD is significantly lower than that of CT. Normal chest radiographs at initial presentation have been reported in approximately 10% to 42% of children who were later confirmed to have ILD. In clinical practice, it is uncommon to proceed to CT for suspected chILD without first obtaining a chest radiograph for baseline comparison. Despite its lower resolution, a high-quality chest radiograph provides a readily available, low-radiation option for longitudinal monitoring, avoiding the cumulative exposure associated with repeated CT examinations. For these reasons, chest radiography should be reviewed prior to initial CT assessment and should be considered the first-line modality for follow-up imaging in clinically stable children with ILD.

Given these considerations, the roles of chest radiography in chILD assessment include (1) raising the initial suspicion of ILD, (2) primary evaluation of disease distribution and appearance before CT imaging is available, (3) investigation of extrapulmonary features of specific chILD entities, (4) detecting signs of chronic aspiration (as a primary or exacerbating pathologic condition), and (5) as a follow-up evaluation imaging modality (ideally as part of a routine assessment, similar to the annual evaluations performed in cystic fibrosis care) [76].

Although chest radiography rarely yields a definitive diagnosis of chILD, it may identify conditions that mimic ILD (especially infection), and helps to define the extent and pattern of structural lung alterations [7].

### *3.4. Computed Tomography*

Thoracic CT represents the primary diagnostic modality for identifying and characterizing ILD, demonstrating approximately 91% sensitivity and 71% specificity for differentiating ILD subtypes. Distinct forms of ILD often present with characteristic CT patterns that correlate with underlying histopathology. However, neither CT imaging nor histology alone is sufficient to establish a definitive diagnosis [1]. The most common high-resolution CT (HRCT) feature of ILD is diffuse ground-glass attenuation, while intralobular lines, irregular interlobular septal thickening and honeycombing are observed less commonly. In some young children, large subpleural air cysts in the upper lobes adjacent to areas of ground-glass opacities—interpreted as paraseptal or irregular emphysema—have been also reported [5].

HRCT should be performed in specialized pediatric radiology centers to ensure diagnostic-quality imaging. HRCT shows a strong correlation with histology and has therefore become the reference standard for radiologic assessment. To optimize spatial resolution, thin-section acquisition, minimal field of view, and sharp reconstruction algorithms are generally recommended [5]. CT imaging informs the subsequent diagnostic approach by classifying findings into three categories: 1) chILD present, reliably classifiable, 2) chILD present, but not reliably classifiable, and 3) chILD unlikely [7].

CT accurately identifies and differentiates diffuse, nodular, cystic, bronchiectatic, and atelectatic or consolidative patterns. However, image quality (and interpretability) can be influenced by multiple factors, including scanner manufacturer, protocol settings, and patient preparation (ie, spontaneous breathing, mechanical ventilation, or pressure-gated techniques). Use of phantoms and

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protocols across sites can help reduce variability for some of these factors. Nevertheless, the rapid technological advancements allowing fast scanning without anesthesia in young children introduce additional complexity and variability. A validated CT scoring system for chILD is currently lacking [2].

Imaging findings of hypersensitivity pneumonitis typically progress from ill-defined centrilobular nodularity and ground glass opacification in the acute phase, to more well-defined centrilobular nodularity in the subacute phase, and ultimately progressing to coarse interlobular septal thickening, architectural distortion (fibrosis), and variable ground glass opacification in the chronic phase, often with areas of lobular hypoattenuation [76].

The CT appearance of NEHI is reportedly 100% specific. Patterns of diffuse ground glass opacity and reticulation are reported as common in infants and young children with surfactant genetic disorders [2]. The extent of ground-glass opacification correlates with clinical phenotype, with significantly more ground glass opacification in NEHI patients requiring continuous oxygen compared to those needing only nocturnal oxygen. While specificity is high, the sensitivity reported in the original study was 78% [76].

The typical HRCT features of pulmonary interstitial glycogenosis (PIG) include diffuse ground-glass opacities, interlobular septal thickening, and occasional small cystic changes, reflecting the cellular, non-inflammatory nature of the disease and its predominant interstitial involvement [76].

Imaging findings of lipoid pneumonia are variable, but include extensive, mass-like consolidation, sometimes with internal areas of low attenuation on CT [76].

If less invasive investigations, including CT imaging, fail to identify the underlying cause and clinical severity warrants, lung biopsy may be indicated to achieve a definitive diagnosis [2].

### *3.5. Histology*

Histology has historically been considered the gold standard for diagnosis of chILD and forms the basis of all key classification systems. However, the increasing role of genetic testing is gradually reshaping this paradigm. Although lung biopsy can provide a diagnosis in most cases, it carries important limitations and inherent risks [2]. Surgical lung biopsy is associated with a mortality rate of 1% to 2%. Samples are obtained invasively, cannot be easily repeated, and may not adequately represent diseased regions of the lung. In infants and young children, an open or video-assisted thoracoscopic (VATS) biopsy is usually required, as transbronchial biopsy is generally insufficient. Proper tissue processing is critical, and with relatively few global histopathology experts for these rare conditions, inter-observer diagnostic variability remains a concern [2].

Currently, fewer than 10% of patients with ILD undergo lung biopsy. In many centers, bronchoscopic transbronchial cryobiopsy —a minimally invasive procedure involving rapid freezing of lung tissue—has replaced VATS biopsy for obtaining tissue samples. Cryobiopsy offers a lower complication rate while providing similar diagnostic accuracy [1].

Histopathologic characteristics are not unique to a specific ILD, and overlap exists among different histopathologic and radiologic findings. This is particularly evident in the four most common histopathologic patterns: usual interstitial pneumonia (UIP), nonspecific interstitial pneumonia (NSIP), organizing pneumonia, and diffuse alveolar damage [1].

Regarding the morphological patterns of chILD, the main microscopic and structural findings and their diagnostic implications are summarized below, based on The Lancet Child & Adolescent Health (2019) and Laenger FP et al. [2,7]. This synthesis provides a practical overview of the histopathological characterization of these entities:

- Neuroendocrine cell hyperplasia of infancy (NEHI): inconspicuous architecture of septa, interstitium, and vessels, with an increased number of neuroendocrine cells in ≥ 70% of bronchi, often confirmed by bombesin-positive immunostaining, typically in otherwise normal pulmonary parenchyma.
- Developmental disorders (CAD/AD): pattern resembling canalicular or saccular lung development. Congenital alveolar dysplasia (CAD) shows enlarged, heavy lung with diffuse

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alveolar simplification, widened septa, reduced capillary density, and predominance of type II pneumocytes. Acinar dysplasia (AD) reflects a more severe maturation arrest at the pseudo-glandular or early canalicular phase, with a complete absence of acini and alveoli.

- Chronic neonatal lung disease, chromosomal abnormalities, or pulmonary hypoplasia: enlarged alveoli with reduced in numbers without increased cellularity
- Pulmonary interstitial glycogenosis (PIG): a non-inflammatory interstitial disorder of infancy; diffuse or focal widening of alveolar septa with PAS-positive, glycogen-rich ovoid cells, preserving the alveolar epithelium.
- Alveolocapillary dysplasia with misalignment of pulmonary veins (ACD/MPV): typically presented with severe neonatal respiratory distress; centrally located septal capillaries, distended peribronchial veins, small arterial media hyperplasia, and lymphatic ectasia
- Chronic pneumonitis of infancy (CPI): type II pneumocyte hyperplasia with interstitial edema and focal lymphoid infiltrates; similar patterns may also be observed in surfactant dysfunction, viral infections, or immunodeficiency.
- Lymphocytic interstitial pneumonia (LIP): dense lymphocytic infiltrate with fibrosis and lymphoid aggregates, often associated with autoimmune diseases or immunodeficiency syndromes.
- Nonspecific interstitial pneumonia (NSIP): less dense inflammatory infiltrates with fibrosis and septal thickening, potentially associated with surfactant dysfunction, autoimmune disease, or hypersensitivity pneumonitis.
- Pulmonary alveolar proteinosis (PAP): intra-alveolar accumulation of eosinophilic, granular, cell-poor material containing cholesterol clefts and foamy macrophages, with type II pneumocyte hyperplasia and an otherwise unremarkable interstitium,
- Desquamative interstitial pneumonia (DIP): alveoli filled with foamy macrophages, often linked to surfactant dysfunction, drug reactions, or toxic inhalation.
- Obliterative bronchiolitis: fibrous remodeling and obliteration of distal airways; may result from post-infectious injury, chronic lung allograft dysfunction, or graft-versus-host disease.
- Follicular bronchiolitis/bronchitis: nodular lymphoid infiltrates within bronchiolar walls, often seen in autoimmune disease or common variable immunodeficiency.
- Granulomatous inflammation: variably distribution, with or without necrosis, suggests infection, sarcoidosis, hypersensitivity pneumonitis, vasculitis, or immunodeficiency.
- Surfactant dysfunction disorders: type II pneumocyte hyperplasia with PAS-D–positive intra-alveolar material, intra-alveolar macrophages, septal fibrosis, and abnormal lamellar bodies, often associated with *SFTPB*, *SFTPC*, or *ABCA3* mutations.

### *3.6. MR Imaging*

Although MR imaging has traditionally been limited by low signal intensity, high levels of susceptibility artifact at air-tissue interfaces, and the frequent need for general anesthesia, substantial advances in pulmonary MRI have been achieved in recent years **[**76**]**. MRI is beginning to gain a role alongside CT in thoracic imaging, offering particular value in the follow-up of selected, established diagnoses rather than serving as a primary diagnostic modality for newly suspected child [2].

### *3.7. Ultrasonography*

Ultrasonography is increasingly being adopted as an initial imaging tool for lung assessment, including in adults with idiopathic interstitial lung disease and connective tissue disorders [2]. Although this approach may offer advantages in resource-limited settings, lung ultrasound lacks the ability to differentiate ILD from other conditions that similarly alter the interstitium (eg, pulmonary edema or subsegmental atelectasis). Consequently, it is not an appropriate imaging modality for evaluating suspected chILD **[**76**]**.

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### *3.8. Bronchoalveolar Lavage (BAL)*

The diagnostic value of BAL in chILD has been examined in numerous studies with inconsistent correlation of the findings with specific chILD entities. Its principal value lies in excluding active infection and providing material for microbiological testing. When BAL is performed, samples should undergo standardized morphological assessment, including staining with hematoxylin–eosin, periodic acid–Schiff (PAS), iron, and Sudan. Therefore, although the diagnostic yield of BAL may be limited, if a bronchoscopy is performed, it should be included in the diagnostic work-up [7].

In any case, as reported by Maher [1], the diagnostic process for interstitial lung diseases should always be based on an integrated, multidisciplinary strategy. The accepted approach to ILD diagnosis is multidisciplinary assessment with a team consisting of pulmonologists, radiologists, and, where necessary, pathologists and rheumatologists. This approach emphasizes the need to synthesize clinical features with imaging data and, when indicated, histologic information.

Nathan and colleagues [5] underscore the importance of a structured, stepwise diagnostic process, beginning with a detailed clinical assessment and advancing to imaging, molecular testing, and histopathological evaluation as necessary. The diagnostic pathway should always be guided by patient age, clinical presentation, and disease evolution, with the aim of identifying developmental, inflammatory, or genetic mechanisms underlying the disease process.

## 4. Genetic Diagnosis

### *4.1. Genetic Testing Methodologies*

The diagnostic workup for monogenic forms of interstitial lung disease increasingly relies on next-generation sequencing (NGS). Causative variants can be identified using targeted gene panels, whole-exome sequencing (WES), or whole-genome sequencing (WGS). The choice among these approaches is guided by the patient's clinical presentation, the suspected biological pathway, and the need to detect specific variant types such as deep intronic or structural variants, which are more readily captured by WGS.

To complement sequencing-based diagnostics, chromosomal microarray (CMA) analysis remains useful for detecting pathogenic CNVs that may underlie or contribute to the pulmonary phenotype. This is particularly relevant in disorders such as 5q deletion syndromes or contiguous-gene deletions associated with congenital alveolar dysplasia (CADDS). When somatic mosaicism or low-level clones are a concern (for example in hematologic disease), fluorescence in situ hybridization (FISH) on the appropriate tissue may be complementary.

In the context of suspected telomeropathies, NGS can identify pathogenic variants in genes involved in telomere maintenance (such as *TERT*, *PARN*, *RTEL1* or *DKC1*). However, genetic findings should be integrated with functional assessment of telomere length. Flow-FISH is considered the clinical standard for this purpose and provides confirmation of telomere shortening across specific leukocyte subsets, thereby supporting the molecular diagnosis and refining phenotype interpretation.

### *4.2. The Importance of Genetic Counselling in Clinical Practice*

Based on the findings of this literature review, defining a structured diagnostic pathway under the guidance of a multidisciplinary team is essential to determine the most appropriate genetic test for each patient. Establishing a well-founded diagnostic suspicion not only optimizes the selection of molecular analyses but also reduces unnecessary expenditure of time and resources in identifying the underlying etiology.

Once a molecular diagnosis is established, genetic counseling plays a pivotal role in providing patients and their families with clear information regarding the disease mechanism, prognosis, and recurrence risk. Counseling allows for informed decision-making and supports families in understanding the implications of the genetic findings.

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For autosomal recessive disorders, parents should be informed that they may be healthy carriers and that each subsequent pregnancy carries a 25% recurrence risk, independent of fetal sex. Counseling should also address the availability of prenatal diagnosis and the possibility of medically assisted procreation (MAP) to manage reproductive risk.

For autosomal dominant conditions, it is important to determine whether the identified variant is inherited or de novo. Segregation analysis in the parents clarifies carrier status and helps evaluate the potential contribution of incomplete penetrance. These considerations are particularly important when interpreting variants of uncertain significance (VUS), as understanding familial segregation can provide critical evidence toward reclassification.

Overall, integrating genetic counselling into clinical practice ensures comprehensive family guidance, supports accurate risk assessment, and facilitates informed choices regarding management, surveillance, and reproductive planning.

## 5. Therapeutic Implications

### *5.1. Precision Medicine and Targeted Therapies*

The management of ILDs has shifted from empirical, phenotype-based approaches to mechanism-driven strategies grounded in molecular genetics and disease-specific pathobiology. The chILD-EU consortium and subsequent European and North American statements have established a framework that integrates genetic, radiologic, and clinical data to guide diagnostic and therapeutic decisions, emphasizing multidisciplinary assessment referral to specialized centers, and biologically informed treatment strategies [3,5,75,77]. Within this framework, supportive measures such as oxygen supplementation, nutritional optimization, infection control, and prevention of pulmonary hypertension, remain the therapeutic cornerstone. Pharmacologic interventions are increasingly tailored to the dominant pathogenic mechanism rather than applied uniformly across diagnostic categories [7].

For inflammatory or organizing forms of chILD, ERS and ATS recommendations support systemic corticosteroids as first-line agents, followed when necessary by steroid-sparing immunosuppressants such as mycophenolate mofetil or azathioprine [7,75]. In immune-mediated and connective-tissue–disease–associated ILD, biologic and targeted immunomodulatory therapies are increasingly employed. Rituximab, abatacept, tocilizumab, belimumab, and other pathway-specific agents have shown encouraging results in pediatric cohorts, improving pulmonary function and facilitating glucocorticoid tapering, particularly in juvenile dermatomyositis–associated ILD and overlap syndromes [78]. Janus kinase (JAK) inhibitors such as ruxolitinib and baricitinib have become key therapeutic options for interferon-mediated and immune-driven ILDs, including COPA syndrome, SAVI, and TREX1- or IFIH1-related interferonopathies, by attenuating aberrant JAK–STAT signaling and type I interferon responses, with improvements in systemic inflammation, digital vasculopathy, and lung imaging [3,78]. These therapies require close hematologic and infectious monitoring, and interferon-stimulated gene expression profiling is increasingly used as a biomarker of therapeutic response.

Surfactant metabolism disorders exemplify the shift toward genotype-based management. Variants in *SFTPB*, *SFTPC*, and *ABCA3* represent the most frequent monogenic causes of neonatal or early-onset ILD, with clinical severity ranging from lethal neonatal respiratory failure to chronic fibrosing disease [7,14]. In *SFTPB* deficiency, where surfactant absence is irreversible, lung transplantation is the only curative option and should be considered early. In contrast, partially functional *SFTPC* and *ABCA3* variants may allow a period of medical stabilization, although treatment responses are highly variable and largely variant-specific. Recent series indicate that corticosteroids and hydroxychloroquine (HCQ) often provide only modest or transient benefit, particularly in misfolding-dominant SFTPC disease, where epithelial injury and endoplasmic-reticulum stress rather than reversible inflammation drive progression [10,12]. Case-based evidence also highlights mutation-specific HCQ efficacy: in an infant with a novel *SFTPC* variant (c.325–

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47_374del), early HCQ treatment failed to prevent fatal respiratory failure, underscoring that misfolding mechanisms may not be responsive to HCQ. Current expert opinion therefore recommends restricting HCQ to short, closely monitored trials and discontinuing it in the absence of early clinical improvement [79].

Telomere biology disorders, like other genetically determined forms of chILD, underscore the value of a mechanism-based therapeutic approach. In telomere-related disease, androgen therapy (e.g., danazol) may stabilize telomere length but carries hepatotoxic and thrombotic risks, necessitating strict monitoring and individualized risk–benefit assessment [80]. Emerging data also indicate that short telomere length in transplant candidates is associated with premature T-cell immunosenescence and increased early rejection risk, with implications for transplant timing and immunosuppressive protocols [80]. In interferonopathies, molecular confirmation of pathway activation is essential before initiating JAK inhibitors, given their toxicity profile and cost [3]. Overall, these developments converge on a unifying principle: in both genetic and immune-mediated pediatric ILD, early molecular and immunologic profiling is crucial to discriminate structural, misfolding-dominant, and immune-driven phenotypes, prioritize supportive care, avoid prolonged ineffective corticosteroid or cytotoxic exposure, and identify candidates for targeted biologic, antifibrotic, or transplantation strategies [7,81].

### *5.2. Impact of Genetic Diagnosis on Management Decisions*

Genetic testing has fundamentally reshaped the management of pediatric ILDs by improving diagnostic precision, refining prognostic assessment, and informing therapeutic choices. Identification of a causal variant allows clinicians to distinguish inflammatory phenotypes, which may respond to immunomodulatory therapy, from structural or misfolding-related defects in anti-inflammatory agents are typically ineffective. In surfactant dysfunction disorders, genotype strongly influences both disease trajectory and treatment strategy: *SFTPB* loss-of-function variants mandate early referral for lung transplantation, whereas many *SFTPC* and *ABCA3* variants exhibit more heterogeneous courses and may justify time-limited trials of medical therapy before irreversible fibrotic remodeling occurs [7]. A recent case report of a compound heterozygous *ABCA3* mutation in a neonate with severe respiratory distress underscored how early molecular diagnosis can confirm a structural etiology, prevent futile escalation of corticosteroids or immunosuppressants, and support timely transplant planning [82].

Genetic information is equally critical in telomere biology disorders, in which pathogenic variants influence therapeutic decisions, transplant conditioning regimens, and long-term monitoring. Short telomere length and bone-marrow dysfunction increase the risk of hematologic toxicity, infection, and post-transplant complications, necessitating adapted conditioning protocols and careful immunosuppressive strategies [80]. Similarly, in interferonopathies such as COPA syndrome and STING1-associated disease, molecular confirmation supports the use of JAK inhibitors (ruxolitinib, baricitinib) to suppress the interferon signature and stabilize pulmonary involvement—an approach that would not be appropriate without documented pathway activation [3].

The first randomized, placebo-controlled phase 2 trial of HCQ in genetically confirmed chILD further underscores the importance of genotype-guided decision-making. In this chILD-EU multicenter crossover study, which included children carrying variants in *SFTPC, ABCA3, NKX2-1, TBX4*, and *COPA*, HCQ was well tolerated but did not significantly improve oxygen requirement, clinical respiratory scores, or growth compared with placebo, suggesting a limited and mechanism-specific benefit [83]. These findings support expert recommendations that HCQ and corticosteroids should be reserved for phenotypes with clear inflammatory components and used only in time-limited trials with predefined response criteria. They also reinforce the need for early genetic testing to avoid prolonged empirical therapy [84]. Neonatal management guidelines and national consensus statements echo this paradigm: in neonates with diffuse interstitial lung disease of suspected genetic origin, supportive care and exclusion of infectious or cardiac causes are prioritized, whereas

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prolonged corticosteroid or HCQ use is discouraged in the absence of demonstrable inflammation. Early molecular diagnosis and timely referral for transplantation are strongly encouraged [85].

Beyond therapy selection, genetic results guide longitudinal management, including timing of transplant referral, eligibility for targeted therapies, and surveillance strategies. Progressive fibrosing patterns, persistent oxygen dependence, or a forced vital capacity decline greater than 10% per year despite optimized care should prompt early multidisciplinary evaluation in transplant centers, particularly when high-risk genotypes or telomere-related defects are present [7]. Psychosocial support, family counseling, and structured transition programs for adolescents with chronic ILD integral components of this genotype-guided model of care [62,86]. Overall, the integration of genetic data into routine practice has transformed pediatric ILD from a largely empirical field to a precision-medicine discipline in which genotype-driven decision-making, multidisciplinary coordination, and proactive monitoring define the current standard of care [86].

### *5.3. Emerging Therapies and Clinical Trials*

The therapeutic landscape of pediatric ILD is rapidly evolving, with antifibrotic agents, targeted immunomodulators, and experimental molecular therapies collectively redefining management paradigms. The most substantive advance to date is the introduction of antifibrotic therapy. The InPedILD trial demonstrated that nintedanib, a multitarget tyrosine-kinase inhibitor acting on VEGF, PDGF, and FGFR pathways, significantly reduced the rate of lung-function decline in children ≥ 6 years with progressive fibrosing ILD, with a safety profile comparable to that observed in adults [77]. Subsequent analyses and expert reviews have positioned nintedanib as the first disease-modifying drug for progressive pediatric ILDs regardless of genetic background, while highlighting the need for long-term surveillance of growth and hepatic function and the potential role of pirfenidone in mixed inflammatory–fibrotic phenotypes or in combination regimens [3,62].

Parallel progress in immune-mediated ILD has been driven by biologics and small-molecule inhibitors. Biologic agents targeting B- and T-cell pathways—rituximab, abatacept, tocilizumab, belimumab—have shown clinically meaningful improvements in pediatric connective-tissue–disease–associated ILD and overlap syndromes, enabling steroid tapering and stabilization of lung function [78]. For interferonopathies and autoinflammatory ILDs, JAK inhibitors provide a pathway-specific approach; pediatric series report improved systemic inflammation, better oxygenation, and radiologic stability with ruxolitinib or baricitinib, often combined with gradual glucocorticoid reduction. Dynamic monitoring of interferon-stimulated gene signatures is emerging as a tool for dose adjustment and early identification of responders [3,87]. These agents exemplify the shift from empirical immunosuppression toward mechanism-based immunomodulation in pediatric ILD. Beyond pharmacologic modulation of inflammation and fibrosis, molecular and gene-based approaches aim to correct or compensate for underlying genetic defects. In surfactant dysfunction disorders, preclinical and early translational studies have identified chemical chaperones (e.g., 4-phenylbutyrate, carbamazepine) and autophagy enhancers (e.g., trehalose, rapamycin) as potential approaches to mitigate misfolding-induced cellular stress and restore proteostasis [77,88]. Patient-derived induced pluripotent stem cells (iPSCs) and lung organoids now allow functional testing of these compounds and evaluation of vector-based correction strategies in mutation-specific models [7]. Over the longer term, gene-editing and RNA-based therapies—including CRISPR/Cas9, base and prime editing, antisense oligonucleotides, and mRNA replacement—offer attractive possibilities for monogenic ILDs caused by *SFTPC*, *ABCA3*, or *SFTPB* variants. However, challenges related to delivery, safety, and long-term genomic stability currently confine these approaches to experimental settings [7,86]. International research networks and registries are critical to translating these advances into clinical benefit. The EU-funded “Orphans Unite: chILD Better Together” initiative created the first international registry and biobank dedicated to pediatric ILD, standardizing diagnostic protocols, centralizing data collection, and enabling multicenter trials [89]. The chILD-EU platform, together with ERS Clinical Research Collaborations, now underpins multi-omics integration, biobank-linked translational studies, and early-phase trials testing targeted therapies [5,77,86]. In

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summary, antifibrotic, immunomodulatory, and emerging gene-based therapies are progressively shifting pediatric ILD management away from predominantly supportive management toward disease-modifying, mechanism-based interventions defining a translational roadmap in which pediatric ILD serves as a model for precision therapeutics in rare respiratory disorders.

## 6. Impact of Genetic Findings on Prognosis

Genetic discoveries have fundamentally reshaped the diagnostic and prognostic landscape chILD. Whereas these disorders were historically defined by clinical and radiologic features alone, molecular characterization now enables increasingly accurate predictions regarding disease severity, trajectory, and long-term outcomes. Understanding the genetic basis of chILD informs expectations for progression, therapeutic responsiveness, and extra-pulmonary involvement, and it supports anticipatory guidance for families, targeted monitoring strategies, and reproductive counseling.

Genetic testing has become a key tool for anticipating disease course. Beyond identifying the underlying pathogenic mechanism, the genotype often determines expected severity, clinical trajectory, and treatment responsiveness. The following examples illustrate how genetic characterization informs prognostic assessment across distinct forms of chILD. Robust genotype–phenotype correlations are most clearly established among surfactant dysfunction disorders. Although many *SFTPC* variants are associated with relatively mild disease and favorable long-term survival, outcomes vary considerably with age at onset and variant type. In a retrospective analysis of 22 genetically confirmed cases (11 *SFTPC*, 5 *ABCA3*, 6 *NKX2-1*), important prognostic differences emerged [90]. Children with *NKX2-1* mutations had the earliest onset (median 0.4 years), the highest baseline respiratory burden, and the greatest prevalence of pulmonary hypertension (66.7%). In contrast, SFTPC-associated ILD presented with milder symptoms and no pulmonary hypertension. Serum KL-6 levels paralleled these patterns. Longitudinal outcomes diverged further: patients with *NKX2-1* variants experienced progressive respiratory decline, whereas those with *SFTPC* variants showed clinical, radiologic, and biomarker improvement. Mortality also reflected this gradient, with no deaths in the SFTPC cohort and 60% mortality in the *NKX2-1* group despite corticosteroid–hydroxychloroquine therapy.

Additional cohort analyses reinforce the heterogeneity of SFTPC-related disease. In an Argentine series of 14 children, neonatal respiratory distress was more frequent and more severe in ABCA3-related disease, while *SFTPC* variants generally conferred more favorable long-term outcomes [9]. Similarly, a Japanese cohort of 20 patients showed that neonatal-onset pulmonary alveolar proteinosis and specific high-risk variants (e.g., p.Leu45Arg or exon 4 splicing mutations) predict poor prognosis [13]. Collectively, these studies highlight the broad spectrum of SFTPC-associated ILD and the importance of variant-level risk stratification in prognostic assessment.

Although extremely rare, surfactant protein-B (SP-B) deficiency represents the most severe end of the surfactant dysfunction continuum. In a series of 11 genetically confirmed cases, complete loss-of-function variants caused fatal neonatal respiratory failure, whereas hypomorphic alleles permitting minimal residual SP-B expression allowed survival beyond infancy [91]. This contrast underscores how even small differences in protein function profoundly alter clinical trajectory.

The *ABCA3* genotype is a major determinant of prognosis, with clinical severity largely dependent on the degree of residual protein function. In a landmark cohort of 185 children, null/null genotypes caused severe neonatal respiratory failure with near-universal early mortality, whereas genotypes retaining partial function (null/other or other/other) were associated with variable presentation and significantly improved early survival [14]. Long-term data from a register-based cohort of 44 children surviving beyond the first year of life demonstrated that missense and small indel variants typically lead to chronic progressive ILD, though many patients survive into later childhood; absence of oxygen requirement was associated with longer survival [17]. These observations highlight the combined prognostic influence of variant type and early disease severity.

Variants in *FLNA* are increasingly recognized as causes of severe early-onset ILD. In a cohort of nine children, all required early respiratory support, three died in infancy from refractory respiratory

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failure and pulmonary hypertension, and survivors demonstrated persistent obstructive physiology with recurrent or ongoing pulmonary hypertension during follow-up [92]. Although some patients stabilize, FLNA-related ILD carries substantial early mortality and chronic cardiopulmonary morbidity.

Monogenic immune dysregulation disorders may present with progressive ILD, especially in late childhood or adolescence. These patients often have complex multisystem inflammatory phenotypes but lack a unifying diagnosis until genetic testing is pursued. Molecular confirmation is essential, as several conditions—such as SAVI—are amenable to precision therapies. In SAVI, initiation of JAK inhibitor therapy can halt or reverse ILD progression, demonstrating how early genetic identification directly improves prognosis [93].

## 7. Future Directions

### *7.1. Knowledge Gaps and Ongoing Research*

Despite substantial progress in the classification and molecular characterization of pediatric ILDs, important gaps persist across diagnosis, pathobiology, and therapeutic evidence. The current ERS and ATS frameworks integrate clinical, radiologic, genetic, and pathological features, yet many children present with overlapping or atypical phenotypes that do not fit neatly into existing categories, and these schemes have not been rigorously validated in large, prospective pediatric cohorts [84]. Genotype–phenotype correlations for key surfactant- and transcription factor–related genes (e.g., *SFTPC, ABCA3, NKX2-1, TBX4*) remain incomplete, with considerable inter-individual variability in age of onset, radiologic pattern, and response to therapy, complicating prognostication and individualized treatment planning [88].

Radiologic assessment is limited by the absence of standardized HRCT scoring systems tailored to developmental anatomy and pediatric ILD patterns. Quantitative imaging and AI-based approaches show promise for objective assessment of disease burden and progression, these methods remain exploratory and unvalidated across centers [75]. Biomarker research is similarly underdeveloped: candidate markers such as KL-6, SP-D, and selected cytokine signatures (e.g., IL-17A, TGF-β) have been associated with disease activity and severity, but none have reached widespread clinical use due to small sample sizes, heterogeneous methodologies, and lack of longitudinal validation [75].

Therapeutic evidence remains a major unmet need. Most treatment strategies in pediatric ILD are extrapolated from adult data or rely on case series and retrospective cohorts, with limited randomized or prospective trials available to guide optimal drug selection, dosing, treatment duration, and monitoring protocols [7]. International registries have begun to clarify natural history and outcomes, but large-scale, disease-specific intervention studies are still lacking. Addressing these gaps will require sustained multicenter collaboration, standardized data collection, and integration of mechanistic research with clinical trial design.

### *7.2. Potential of Multi-Omics and Gene Editing*

Multi-omics approaches offer an unprecedented opportunity to overcome many of the current limitations in pediatric ILD. While genomic sequencing has already transformed diagnosis, particularly for surfactant dysfunction and other monogenic disorders, the addition of transcriptomic, proteomic, metabolomic, and epigenomic data is beginning to delineate disease-specific stress responses, immune signatures, and fibrotic pathways that cannot be fully appreciated through imaging or histopathology alone [94].

Patient-derived iPSCs and lung organoids provide complementary functional platforms in which patient-specific mutations can be modeled and candidate therapies—such as autophagy modulators, chaperone-based treatments, and antifibrotic agents—can be tested under controlled conditions [95]. These systems enable rapid assessment of drug toxicity and efficacy, facilitate the study of rare genotypes, and offer a bridge between bench discoveries and early-phase clinical trials.

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Gene editing and RNA-based therapies represent the most ambitious frontier. Technologies such as CRISPR/Cas9, base editing, and prime editing offer proof-of-concept for correcting pathogenic variants in surfactant and telomere-related genes, while antisense oligonucleotides and mRNA replacement therapies may allow transient or partial functional rescue [77]. However, major challenges remain, including efficient and safe delivery to the distal airways, avoidance of off-target effects, long-term genomic stability, and ethical considerations surrounding germline and pediatric interventions. For the foreseeable future, these approaches are likely to remain confined to experimental models and highly selected early-phase trials, but they nonetheless provide a conceptual framework for curative strategies in monogenic pediatric ILDs [7].

### *7.3. Personalized Medicine in Pediatric ILDs*

Personalized medicine is emerging as the future paradigm of pediatric ILD care, integrating genetic, molecular, radiologic, physiologic, and digital-health data into individualized management plans. Genotype-guided strategies in surfactant dysfunction disorders illustrate this need: variability in clinical presentation and trajectories among *SFTPC*, *ABCA3*, and *NKX2-1* variants—well documented across international cohorts—highlights the importance of precise molecular diagnosis and early stratification [90]. Misfolding-prone *SFTPC* variants may benefit from chaperone or autophagy-targeted therapies, whereas *ABCA3* loss-of-function variants often progress to fibrosis, warranting earlier consideration of immunomodulatory or antifibrotic agents and timely evaluation for transplantation [88].

Achieving truly personalized medicine will depend on the convergence of three elements: (1) routine genome-wide diagnostics with rapid turnaround for suspected chILD; (2) interoperable international registries integrating longitudinal clinical and multi-omic datasets; and (3) translational platforms such as patient-derived organoids, iPSC-derived AT2 cells, and in-vivo models that enable functional validation and preclinical drug screening [86]. These components collectively support biomarker discovery, adaptive trial designs, and the transition of monogenic chILD from supportive management to targeted or disease-modifying therapies. Integrated multimodal data streams—including biomarkers, quantitative HRCT metrics, home spirometry, wearable sensors, and remote auscultation—may further enable early detection of exacerbations and more precise therapy titration, while psychosocial factors remain essential for shared decision-making [62].

International collaboration is essential. The European chILD registry and similar initiatives provide platforms for biomarker validation, refinement of classification systems, and the development of genotype- and pathway-specific trials [5,86,89]. The integration of multi-omics, advanced imaging, digital health technologies, and multinational registries is expected to embed personalized medicine into routine care and advance pediatric ILD toward a truly mechanism-driven discipline.

## 8. Conclusions

Genetic testing plays a central role in the management of chILD, linking clinical expression to the underlying biology. Molecular diagnosis enables accurate prognostic stratification, supports individualized follow-up, and informs targeted therapeutic strategies within a precision-medicine framework. A confirmed genetic diagnosis is also essential for providing families with accurate recurrence-risk counseling and for planning timely care in future pregnancies. Furthermore, integrating genomic testing with multi-omics approaches, functional models, and international registries paves the way toward predictive and personalized medicine. Early integration of comprehensive molecular diagnostics into clinical evaluation is therefore crucial to refine disease classification, bridge current knowledge gaps, and advance the management of pediatric ILD toward an etiological mechanism-driven framework.

**Supplementary Materials:** The following supporting information can be downloaded at the website of this paper posted on Preprints.org.

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## Abbreviations

The following abbreviations are used in this manuscript:

| | |
|---|---|
| chILDs | Children's interstitial lung diseases |
| CTD-ILD | Connective tissue disease–associated ILD |
| NEHI | Neuroendocrine cell hyperplasia of infancy |
| DIP | Desquamative interstitial pneumonia |
| NSIP | Non-specific interstitial pneumonia |
| HRCT | High-Resolution Computed Tomography |
| IPF | Idiopathic pulmonary fibrosis |
| VATS | Video-assisted thoracoscopic surgery |
| MDD | Multidisciplinary diagnostic discussions |
| SPC | Pulmonary-associated surfactant protein C |
| SPB | Surfactant protein B |
| snoRNP | small nucleolar ribonucleoprotein |
| SAVI | STING-associated vasculopathy with onset in infancy |
| COPI | Coatomer protein complex I |
| GH | Growth hormone |
| GRAF | GTPase Regulator Associated with Focal Adhesion Kinase |
| FINCA | FIbrosis, Neurodegeneration and Cerebral Angiomatosis |
| CNVs | Copy Number Variations |
| PIG | Pulmonary interstitial glycogenosis |
| CT | Computed Tomography |
| VATS | Video-assisted thoracoscopic |
| UIP | Usual interstitial pneumonia |
| NSIP | Nonspecific interstitial pneumonia |
| CAD | Congenital alveolar dysplasia |
| AD | Acinar dysplasia |
| ACD | Alveolocapillary dysplasia |
| MPV | Misalignment of pulmonary veins |
| CPI | Chronic pneumonitis of infancy |
| LIP | Lymphocytic interstitial pneumonia |
| NSIP | Nonspecific interstitial pneumonia |
| PAP | Pulmonary alveolar proteinosis |
| DIP | Desquamative interstitial pneumonia |
| MRI | Magnetic resonance imaging |
| PAS | Periodic acid–Schiff |
| BAL | Bronchoalveolar lavage |
| NGS | Next-generation sequencing |
| WES | Whole-exome sequencing |
| WGS | Whole-genome sequencing |

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| | |
|---|---|
| CMA | Chromosomal microarray |
| FISH | Fluorescence in situ hybridization |
| CADDS | Contiguous ABCD1/DXS1357E deletion syndrome |
| VUS | Variants of uncertain significance |
| ERS | European Respiratory Society |
| ATS | American Thoracic Society |

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